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Title: Biomedical Applications of Microfluidic Technology

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Biomedical Applications of Microfluidic Technology

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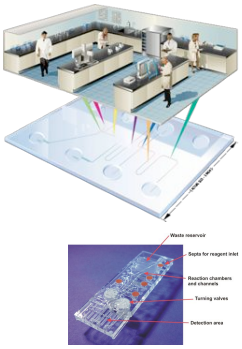
What are the microfluidic lab-on-chips?

- **Lab-on-chips** :
  A class of submillimetre size bioanalytical devices.
- **Perform**:
  fluidic processes,
  sensing, analysis and separation of biochemical samples.
- **Integrate**:
  fluidics, electronics, optics and biosensors.
- **Analyse**:
  metabolites, molecules, proteins, nucleic acids, cells and viruses.



Advantages and Applications

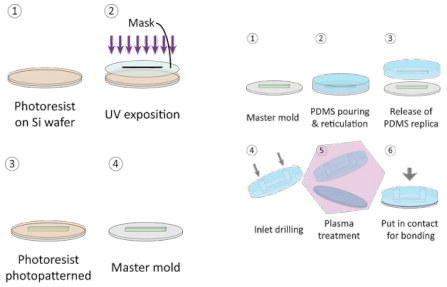
- Low fluid consumption (less waste, lower reagents costs and less required sample volumes for diagnostics)
- Faster analysis and response times due to short diffusion distances, fast heating, high surface to volume ratios, small heat capacities.
- Better process control because of a faster response of the system (e.g. thermal control for exothermic chemical reactions)
- Small footprint
- Massive parallelization allows high-throughput analysis
- Lower fabrication costs allow mass production
- Safer platform for chemical, radioactive or biological studies because of integration of functionality, smaller fluid volumes and stored energies

- PCR
- Immunoassays
- drug screening
- cell biology
- Genomic & proteomics
- single cell manipulation

- Water & food quality
- Environmental monitoring

- Improvements in synthetic yields
- Novel reactions by control surface chemistry, local heat and mass transfer
- Controlling reagent concentrations

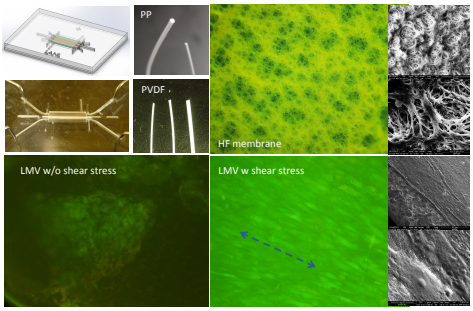
Microfluidic Device Fabrication Using Polydimethylsiloxane (*PDMS*)



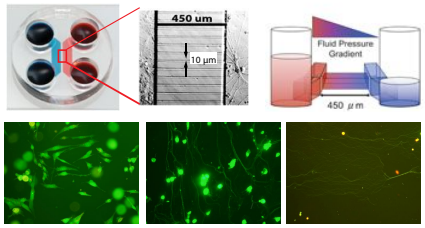
Microfluidic Applications at Bioscience Current status

- Tissue engineering
  - Liver tissue/vascular tissue engineering
  - Control neuron differentiation
- Droplet-based microfluidic Technology
  - Cell and bacteria encapsulation in W/O droplet
  - Algae encapsulation in agar droplet
  - Double emulsion (W/O/W droplets)
- Applications
  - Digital PCR
  - Protein crystalization
- Future directions

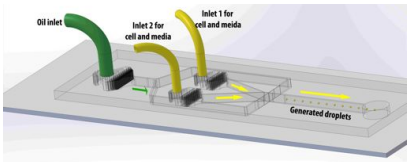
Microfluidic studies Develop Liver Microvascular Tissue

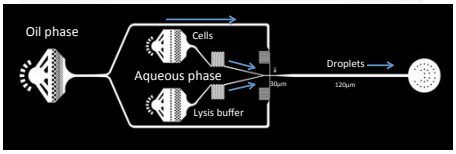


Neuron Microfluidic Device



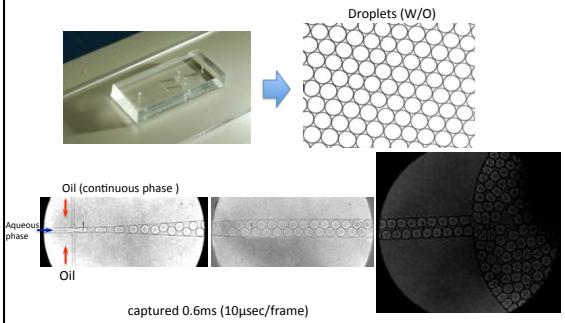
Droplet-based Microfluidic Technology





The combination of aqueous phase is flexible. It can be chemical reaction solution, cell culture media with cells, agarose/beads and other buffers.

Microfluidic droplet generator (W/O)



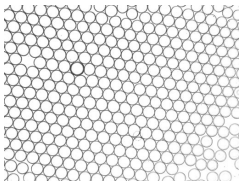
captured 0.6ms (10µsec/frame)

Real-time Changing the Size of Droplet (50-480ul/h)

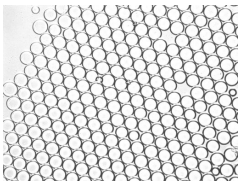
Real-time changing the droplet size: The droplet will contain different amount of reaction solution for reaction efficiency evaluation.

Stability Evaluation

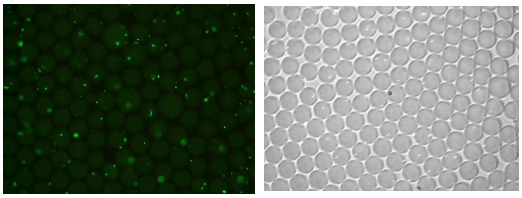
1 hr after generation



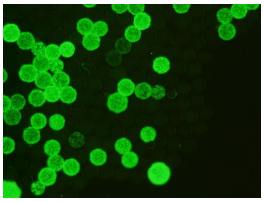
After 24 hr in 4°C

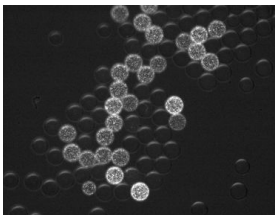


Single Bacteria Encapsulation BL21 (DE3) Gold (pETck3)

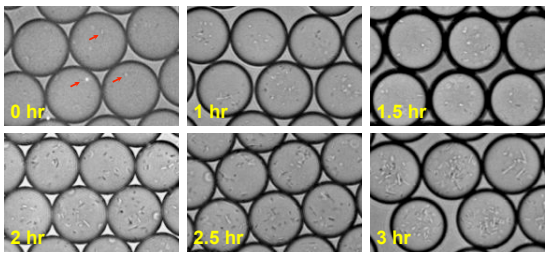


BL21 (DE3) Gold (pETck3) (Overnight incubation @ 37°C)

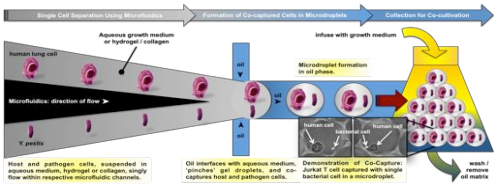


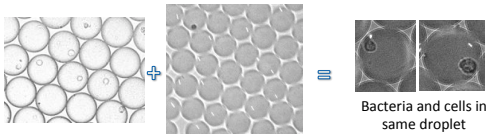


E. Coli Proliferation in W/O Droplets



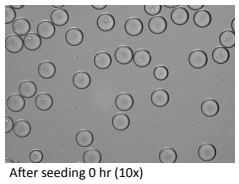
Cell and Bacteria Coencapsulation



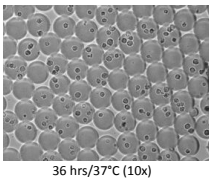


Bacteria and cells in same droplet

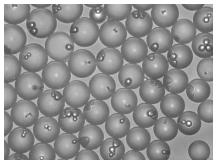
Encapsulated Human Jurkat T Cells



After seeding 0 hr (10x)

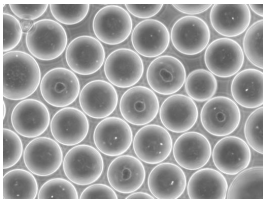


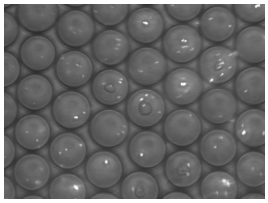
36 hrs/37°C (10x)



Over two weeks at 4°C (20x)

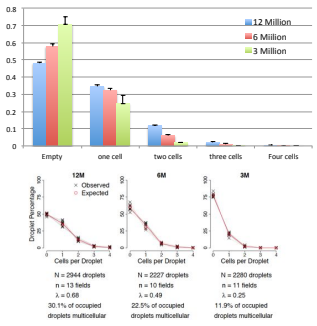
Phagocytosis (THP1 and *E. Coli*)



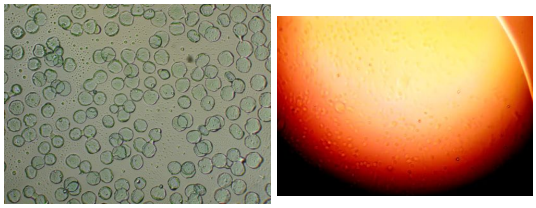


Encapsulation Efficiency

(Poisson Distribution)

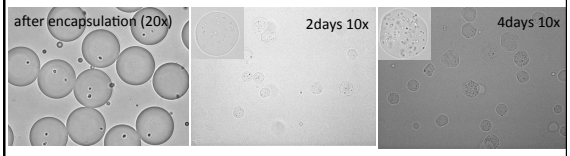


Move from Aqueous Droplet to Hydrogel Droplet

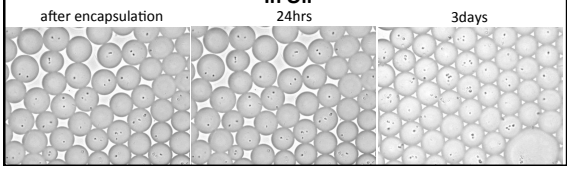


Nannochloropsis in Agar Droplets

In F2 media

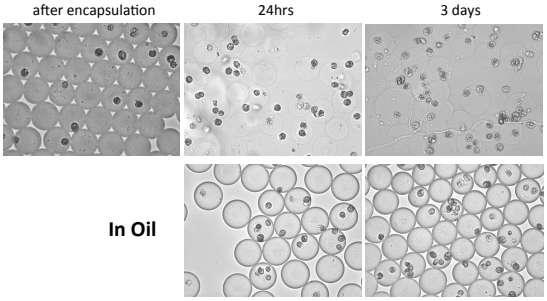


In Oil

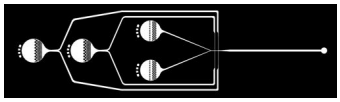


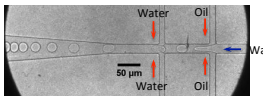
Tetraselmis in Agar Droplet

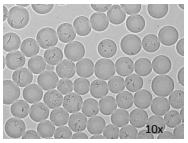
In F2 media

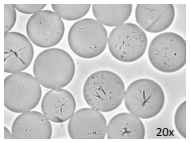


Double Emulsion (W/O/W)

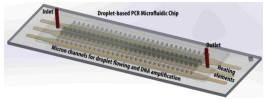


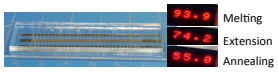


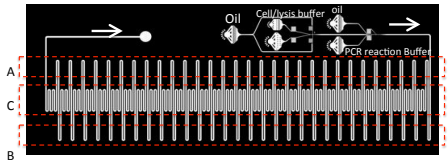




Microfluidic PCR







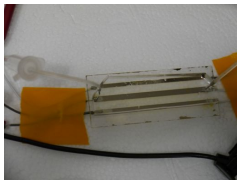
$$t_i = \frac{l_i b_i \delta_i}{Q}$$

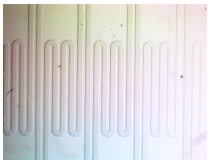
t_A:t_B:t_C=4:4:8.94

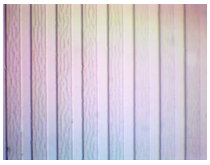
Residence times of PCR reagent are in the ratio 4:4:9 for respective zones melting, extension and annealing

Long channel: 10mm; short channel: 2.5mm

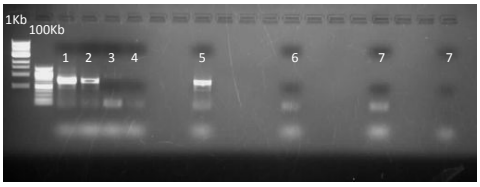
Microfluidic PCR







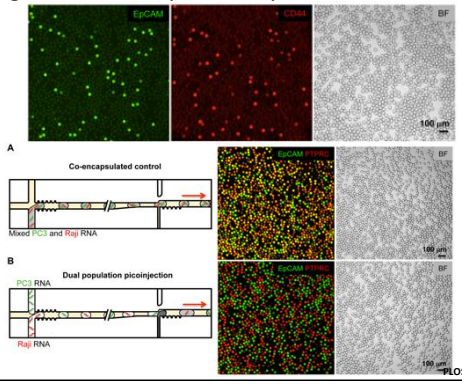
mAb PCR Gel



1. Regular PCR
2. Regular PCR using droplets
3. Regular PCR w/o template
4. Regular PCR w/o template using droplets
5. Microfluidic PCR w/flow rate 20µl/hr
6. Microfluidic PCR w/o template 20µl/hr
7. Microfluidic PCR w/flow rate 200µl/hr
8. Microfluidic PCR w/flow rate 200µl/hr (leftover)

Sample loading in gel: 3µl + 3µl dye
Reaction: 2.5µL template was used in 50µL (total volume)

Digital RT-PCR Taqman assays in microfluidic droplets



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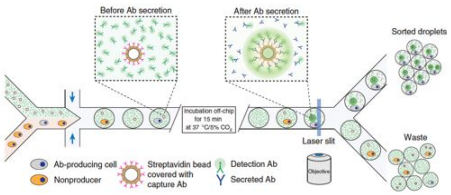
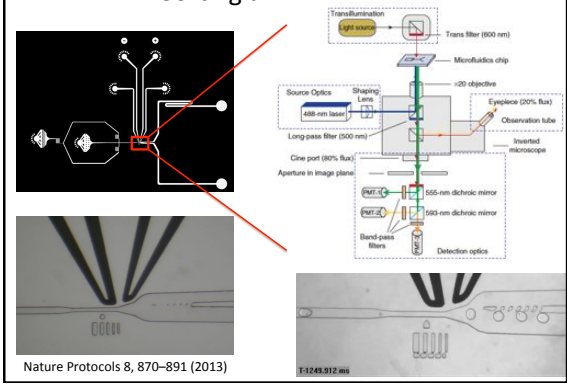
Principle of cell sorting



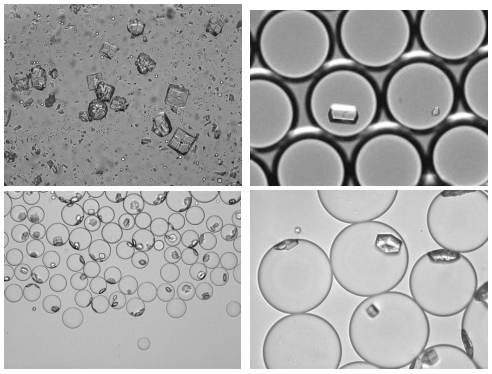
Nature Protocols 8, 870–891 (2013)

Sorting and Detection



Nature Protocols 8, 870–891 (2013)

Lysozyme crystallization in droplet (20hrs)



Future

- Multiplexing
- Not just pathogen detection, but detection of virulence and antibiotic resistance markers
- Organism sensing and detections of biomarkers for infection together
- Point of care application in resource limited setting

Acknowledgment

- Dr. Andrew Bradbury and his team
- Dr. Elizabeth-Hong Geller
- Dr. Scott Twary
- Dr. Jeri Sullivan